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Inside JEB

PAINFUL FEELINGS IN CRABS

Robert Elwood

Have you ever wondered, whilst tucking into a tasty prawn cocktail, whether those delicious little crustaceans ever felt pain? When Professor Robert Elwood was asked this very same question by a well-respected British seafood chef, Elwood found he didn't have a definitive answer. Having previously studied many aspects of crustaceans' lives Elwood pondered this question for a few weeks before beginning his investigations, using the common shore crab as his subjects to tackle the question (p. 353).

From the outset, Elwood and his student, Barry Magee, wanted to carefully design an experiment that would distinguish between pain and a phenomenon known as nociception. Explaining the difference, Elwood says, 'The function of pain is to aid future avoidance of the [painful] stimulus, whereas nociception enables a reflex response that provides immediate protection but no awareness or long-term behaviour [change]'. While nociception is generally accepted to exist in all animals, the same is not true of pain. Whether pain exists in all animals, but especially in crustaceans, remains widely debated.

Pain results in a learned avoidance of potentially harmful and painful stimuli, but as Elwood notes, it can also lead animals to make trade-offs between keeping a valuable resource and avoiding a painful stimulus. But what resource would a crab value and need convincing to give up? Despite their intimidating appearance with their fearsome pincers, it appears that crabs value dark hideaways beneath rocks where they hunker down to avoid hungry predators. Exploiting the crabs' preference for seclusion, Elwood and Magee tested whether the crustaceans experienced pain by seeing if they could learn to give up a perfect dark hiding place in order to avoid a mild, but potentially painful electric shock.

Capturing 90 crabs at a nearby beach, the duo transported the animals back to their lab at Queen's University, Belfast, UK, ready to test the animals' responses to a gentle electric jolt. Introducing the crabs individually to a tank equipped with two attractive dark shelters, the duo allowed the crustaceans to select their shelter of choice. However, for some of the wannabe lodgers their chosen shelter had an unpleasant surprise in store, dispensing mild electric shocks. Returning the crabs to the tank after a short breather, Elwood and Magee gave them a second chance to sample the shelters, and found that the crabs mostly stuck with what they knew best, returning to shelter that they had chosen first time around, even if it meant receiving another shock. However, when the crustaceans were introduced to the tank for a third time, most of the shocked crabs began exploring alternative options. After just two rounds of shocks the crabs were learning to avoid the shelter where they got zapped – although some crabs (perhaps the unadventurous ones that hadn't found the other welcoming shelter) continued to return to the unfavourable shelter, but this time they were much more likely to scuttle away and give up their precious hideaway to avoid the electric shocks.

Elwood and Magee explain that the crab's swift shock avoidance and discrimination learning (between the two shelters) clearly shows that shock affects their choice of shelter and is consistent with the definition of pain used for other species. The evidence collected from their study thus strongly suggests that crustaceans do indeed feel pain.

10.1242/jeb.084046

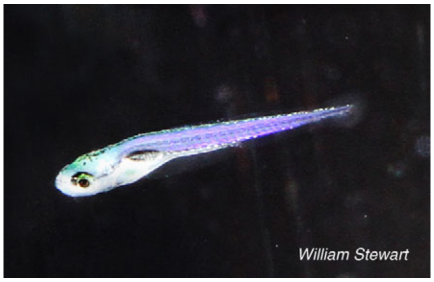
Magee, B. and Elwood, R. W. (2013). Shock avoidance by discrimination learning in the shore crab (*Carcinus maenas*) is consistent with a key criterion for pain. *J. Exp. Biol.* **216**, 353-358.

Nicola Stead

ZEBRAFISH SENSE WATER FLOWS TO AVOID PREDATORS

With a hungry predator on the loose looking for its next meal, you've got to keep your wits about you if you want to survive. But what sensory tools do you employ to detect and escape in time from this peckish hunter? For zebrafish larvae trying to evade their cannibalistic elders, the answer seems to lie in a special sensory system – the lateral line – that detects water motions near the body, explains William Stewart (p. 388).

Whilst watching videos of adult zebrafish snacking on their young, Stewart, a PhD student in Matthew McHenry's lab at the University California, Irvine, USA, noticed that larvae were actually quite effective at dodging these attacks. He realised that the ability to escape was unlikely to rely on the visual system because this system processes information relatively slowly and – given the speed with which some predators strike



– processing speed can be a matter of life or death for the victim. Stewart therefore turned his attention to another sensory system, the lateral line. This sensory system detects water movements or flows over the body and Stewart points out that while 'the system is sensitive to artificially generated water flows, its role in predator evasion is unclear'.

To investigate the role of this sensory system in the survival of onslaughts from adult zebrafish, Stewart enlisted the help of undergraduate student Gilberto Cardenas. Together, they recorded the outcome of predator–prey interactions using larvae in which the sensory system had been removed. To their surprise, larvae without the lateral line system survived a mere 5% of attacks. On the whole, larvae with a functional detection system were 14 times more likely to survive and they survived four times longer than their unfortunate relations without a lateral line system.

To understand the details and precise timings of both the attacks and escape manoeuvres, Stewart turned to videoing the zebrafish larvae–adult encounters with a high-speed, high-magnification video camera. After tackling the problems associated with filming fish that seemed to love swimming rapidly out of the camera's field of view, Stewart was able to determine the exact timing of a larva's escape relative to an incoming attack. The results showed that the larvae responded as soon as the hungry adult started moving and not, as expected, when the adult opened its mouth and started suction feeding (the way in which adults capture larvae). 'We were intrigued that the larvae initiated their response prior to predator suction', says Stewart. He adds, 'The water flows generated by suction are of [a] higher magnitude than the flows produced by the predator's approach'.

It is likely that the movement of the hunter zebrafish, a bit like a ship, creates a bow wave that the larvae sense. Sensing this bow wave, as opposed to water movement caused by suction, allows the targeted larvae more space and time to escape. While other sensory systems, such as vision, may still be important in determining when to perform an emergency escape manoeuvre, Stewart highlights that 'Unlike vision, flow sensing can be effective in both light and dark conditions or when the water is turbid', in addition to being the faster of the two senses. Sensing water movement thus seems to be the best tool in a fish's arsenal to avoid being someone's lunch or dinner, whether it's night or day.

10.1242/jeb.084079

Stewart, W. J., Cardenas, G. S. and McHenry, M. J. (2013). Zebrafish larvae evade predators by sensing water flow. *J. Exp. Biol.* **216**, 388-398.

Nicola Stead

STOCKING UP FOR WINTER: MINKE WHALES' STRATEGIES

Some humans struggle to balance their energy budget in the affluent west, but imagine the complexity of balancing energy intake against expenditure when you gorge for half of the year before embarking on a 6 month fasting migration to your barren equatorial breeding grounds. This is the dilemma faced by minke whales feeding in the abundant waters near Iceland. Fredrik Christiansen from the University of Aberdeen, UK, explains that binging whales store large amounts of excess energy in specialised fat tissue, known as blubber, and scientists are keen to know more about how, when and where mature and immature whales use and store energy. He adds that youngsters are more likely to invest energy in growth, while older (reproducing) whales are more likely to store energy for the lean months ahead, especially if they're pregnant females. Curious to discover more about the whales' energy storage strategy, Christiansen and his colleagues, Gísli Víkingsson, David Lusseau and Marianne Rasmussen decided to analyse the blubber distributions of minke whales in their Icelandic feeding grounds to learn more (p. 427).

Fortunately, Víkingsson, from the Marine Research Institute, Iceland, has more than 20 years of experience of working with whales; he captured over 150 minke whales over a 4 year period, measuring their girth and the thickness of their blubber at 18 different locations across their bodies. Using these measurements, Christiansen was able to calculate the total amount of blubber carried by each animal: 'Measuring absolute blubber volume, rather than the relative blubber thickness, made sense since we wanted to quantify the actual amount of blubber that the whales accumulated, so we could convert this to energy'.

Equipped with his blubber volume estimates, Christiansen was able to calculate the rate of blubber accumulation and the total amount gained throughout the feeding season in whales ranging from fast-growing juveniles to pregnant females and mature bulls. He was then able to ask what specific blubber-gaining strategy different whales used during the summer. Did they pile on the pounds at the very end of their feeding season or did they squirrel energy away gradually? The team revealed that the youngsters didn't gain much blubber – instead they dedicated their energy to growth. However, as expected, pregnant females, who need to stock up for both the migratory journey and motherhood, almost doubled their blubber content to 449 kg, gaining a steady 0.0024 m^3 of blubber each day. Mature males also used the same strategy, steadily piling on an astonishing average of 532 kg, which equates to an impressive 15.5 GJ of energy, enough to keep a 60 W light bulb burning for almost 8 years.

So why do males stock up just as much as females? Do they just enjoy eating or do they really need all this energy? Pondering this question, Christiansen suggests, 'Maybe we underestimate the energetic costs of male reproduction, or reproductive behaviour'. He adds that males may be promiscuous and therefore devote considerable time and energy to searching for mates, while females who expend a lot of energy nursing new-borns may 'benefit from conserving energy by being quite inactive, and instead let the males do the work of locating them'.

It is clear that we still have a lot to learn about these mysterious creatures. However, Christiansen's study will hopefully help us develop non-invasive ways of measuring whale blubber thickness so that we can gain more insight into how they acquire, store and use energy ready for their epic migration south.

10.1242/jeb.084053

Christiansen, F., Víkingsson, G. A., Rasmussen, M. H. and Lusseau, D. (2013). Minke whales maximise energy storage on their feeding grounds. *J. Exp. Biol.* **216**, 427-436.

Nicola Stead

LARGER MIDBRAIN IN MICE MOTIVATED TO RUN

Hitting the treadmill voluntarily everyday is probably not everybody's idea of fun. However, for some house mice – and even humans – exercise seems to be an almost addictive activity. Clearly, these individuals are gifted with astonishing amounts of motivation and/or ability. These attributes, especially motivation, are controlled by the brain and nervous system, and increased brain size has been suggested to increase the ability to undertake and endure exercise. Bigger brains have also been linked with more complex and fascinating behaviour. This all begs the question – does selecting for mice with this intriguing 'gym bunny' behaviour also drive an increase in brain size?

Luckily, for nearly 20 years Theodore Garland and his colleagues at the University of California, Riverside, USA, have been selectively breeding exercise-loving mice and were able to start answering this question (p. 515). However, when the team analysed the total brain mass of these athletic mice they found no change in mass compared with their couch potato relations. Undeterred, the team decided to investigate further. They carefully dissected brains into two different regions, the cerebellum and non-cerebellar areas, and weighed these sections separately. This time, there was a difference. Surprisingly, however, the increase was not in the cerebellum – a region of brain crucial for controlling movement. To pinpoint the exact area in the non-cerebellar regions that was enlarged, the team turned to high resolution imaging to determine the volume of individual components. Interestingly, they found an increase in one specific area – the midbrain region – that occupied up to 13% more volume in the exercise-loving mice.

Whilst the cerebellum is important for co-ordination, the midbrain, as the investigators point out, is also important in controlling various other tasks. This region is essential for reward learning, motivation and reinforcing behaviour. Could it be that these active mice have a heightened reward system that motivates them to exercise? Alternatively, there is evidence that the midbrain is also involved in some aspects of controlling movement. Either way, it is clear that willingness to exercise can evolve from the enlargement of just one specific area of the brain rather than the whole structure.

10.1242/jeb.084061

Kolb, E. M., Rezende, E. L., Holness, L., Radtke, A., Lee, S. K., Obenaus, A. and Garland, T., Jr (2013). Mice selectively bred for voluntary wheel running have larger midbrains: support for the mosaic model of brain evolution. *J. Exp. Biol.* **216**, 515-523.

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